АГРАРЕН УНИВЕРСИТЕТ
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OPINION

on a dissertation for the degree of "Doctor of Science" in: field of higher education 4. Natural Sciences, Mathematics and Informatics, professional field 4.3. Biological Sciences, scientific specialty: Genetics

Author of the dissertation: Prof. Dr. Bojin Maksimov Bojinov, Department of Plant Physiology, Biochemistry and Genetics, Agricultural University - Plovdiv

Topic of the dissertation: "Use of molecular markers in genetic diversity studies and for DNA profiling"

Prepared the opinion: Assoc. Prof. Dr. Lyubka Hristova Koleva-Valkova, Agricultural University - Plovdiv, professional field 4.3. Biological Sciences, scientific specialty Plant Biochemistry, appointed as a member of the scientific jury by Order No. RD 16-753 / 12.06.2025 of the Rector of the Agricultural University - Plovdiv.

1. Relevance of the scientific problem considered in the dissertation

The topic of the dissertation is interesting and relevant. It examines the possibility of selecting and using molecular markers as a means of determining genetic diversity in various organisms (plants, animals, microorganisms), to accelerate the selection process, guarantee genetic identity, and protect intellectual property. The use of DNA-based methods is not new worldwide, but in Bulgaria, it is still poorly used. The traditional reason is the high cost of this type of analysis, which, however, has significantly decreased in recent years. Another significant reason is the insufficient information about the benefits of the applied genetic analyses, the lack of standardization of protocols, etc. The present dissertation manages to fill the gaps in the critical problem considered and to propose solutions for improving selection in Bulgaria. Therefore, I believe that the relevance of the problem in the developed dissertation is high.

2. Aim, hypotheses, tasks, and research methods

The aim of the dissertation work is clearly formulated - to establish a molecular marker system that can be proposed for reliable and reproducible identification of genetic diversity in microbial, plant, and animal organisms. This system can be used for selection purposes and for control in variety testing and seed production in plants, as well as in the breeding of different breeds of animals. In connection with the achievement of this goal, 3 tasks and 4 subtasks have been set. The main hypothesis in the dissertation is that from the many known marker systems, one can be selected that most fully meets the set goal. When performing the tasks, various methods were used, which are described in detail in the "Materials and Methods" section. Molecular marker systems (AFLP, ISSR, CAPS, SSR, RFLP) were used on a large number of biological objects: plants – cotton, tomatoes, tobacco, pepper, wheat, corn, barley, paulownia;

animals – autochthonous goat breeds and fungi – pathogens of the genera *Phytophthora* and *Fusarium*. The results were processed with appropriate methods for statistical analysis and specific software.

3. Visualization and presentation of the obtained results

The dissertation consists of 260 pages and has the necessary parts - Introduction, Literature review, Aim and objectives, Materials and methods, Results, Discussion, Conclusions, Contributions, and References. The cited sources are 280, mainly by foreign authors. The work is illustrated with 17 photos presenting some of the experimental objects used in the study. The results section is voluminous and occupies 95 pages. The main results are presented through 63 figures and 33 tables, which are clear and informative. The results are subjected to statistical analysis with appropriate programs and are considered separately for each of the experimental objects used.

4. Discussion of the results and the literature

In the Discussion section, the results obtained are discussed in depth, both by the type of marker systems used and by the type of biological objects studied, making the necessary comparisons and drawing generalizations valid for the entire group of organisms studied. The arguments presented in the discussion are based on a significant volume of scientific research, which confirms the author's in-depth awareness of the problem under study, as well as his practical experience in the use of molecular markers. The use of the ISSR marker system for identifying genetic diversity is argued, due to the relatively easy methodology and good reproducibility of the results.

5. Contributions of the dissertation work

As a result of the research conducted, the author has derived five theoretical and nine applied contributions, which are clearly formulated and emphasize the significance of the dissertation work.

6. Critical notes and questions

I have no critical notes.

7. Published articles and citations

The author has submitted 9 scientific publications related to the dissertation work and 4 patents for registered cotton varieties. The scientometric assessment of the indicated publications and patents amounts to 254 points, with a minimum required standard of 100. The presented list of citations includes 152 citations of 1 article. Nine of the presented publications received a total of 167 citations. The total number of points from all indicators exceeds the minimum required standard for awarding the scientific degree of Doctor of Sciences.

8. Conclusion

The documents and materials presented by Prof. Bojinov in connection with the procedure for defending a dissertation for the degree of Doctor of Science fully meet the requirements of the Low of the Development of the Academic Staff in the Republic of Bulgaria, the Regulations for the Implementation of the Low in the Republic of Bulgaria and the Regulations for the Implementation of the Low at the Agricultural University - Plovdiv.

The dissertation of Prof. Dr. Bojin Bojinov is an in-depth study on the use of molecular markers for the study of genetic diversity and for DNA profiling in various biological organisms. It uses modern molecular marker systems (AFLP, ISSR, CAPS, SSR and RFLP analyses) and significant scientific results have been obtained, representing an original contribution to science. The dissertation shows that Prof. Bojinov possesses in-depth theoretical knowledge and professional skills in the field of genetics and molecular biology.

Based on the above, I confidently give my positive assessment of the research conducted, the achieved results and contributions, and propose to the esteemed scientific jury to award the scientific degree "Doctor of Sciences" to Prof. Dr. Bojin Maksimov Bojinov in the field of higher education 4. Natural Sciences, Mathematics and Informatics, professional field 4.3. Biological Sciences, scientific specialty "Genetics".

20.08.2025

Prepared by:

Assoc. Prof. Dr. Lyubka Koleva-Valkova